Subterranean Biology 42: 1–22 (2022)
doi: 10.3897/subtbiol.42.75725
https://subtbiol.pensoft.net

RESEARCH ARTICLE

A peer-reviewed open-access journal
Subterranean Biology
Published by The International Society for Subterranean Biology

Unique and fragile diversity emerges from Brazilian caves – two new amphibious species of *Xangoniscus* Campos-Filho, Araujo & Taiti, 2014 (Oniscidea, Styloniscidae) from Serra do Ramalho karst area, state of Bahia, Brazil

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Academic editor: Matthew L. Niemiller | Received 24 September 2021 | Accepted 23 December 2021 | Published 10 January 2022

http://zoobank.org/5C413E74-53FA-4AB0-B4CB-B6A332C83030

Citation: Campos-Filho IS, Gallo JS, Gallão JE, Torres DF, Horta L, Carpio-Díaz YM, López-Orozco CM, Borja-Arrieta R, Aguiar JO, Bichuette ME (2022) Unique and fragile diversity emerges from Brazilian caves – two new amphibious species of *Xangoniscus* Campos-Filho, Araujo & Taiti, 2014 (Oniscidea, Styloniscidae) from Serra do Ramalho karst area, state of Bahia, Brazil. Subterranean Biology 42: 1–22. https://doi.org/10.3897/subtbiol.42.75725

Abstract

Two new troglobitic species of *Xangoniscus* are described from two caves of Serra do Ramalho karst area, Bambuí geomorphological group, state of Bahia. *Xangoniscus lapaensis* **sp. nov.** is described from Gruna Boca da Lapa cave, and *X. loboi* **sp. nov.** from Gruna da Pingueira II cave. Both species are blind and depigmented and show amphibious habits, as observed for all species of *Xangoniscus* described until now. *Xangoniscus lapaensis* **sp. nov.** occurs in travertine pools fed by water of the upper aquifer, and *X. loboi* **sp. nov.** occurs in a small stream, an upper vadose tributary. Both species occur in fragile microhabitats. Ecological and behavioral data, conservation remarks, and IUCN conservation assessments are included to provide background data for conservation efforts in this unique karst area.

Keywords

Bambuí geomorphological group, Limestone, Terrestrial isopods, Troglobitic, Upper Aquifer

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Introduction

Terrestrial isopods (Oniscidea) are the only crustacean lineage completely adapted to the terrestrial way of life (Hornung 2011; Richardson and Araujo 2015; Taiti 2018). The group occurs in almost all kinds of terrestrial habitats, including caves (Schmalfuss 2003; Taiti 2004). Oniscidea comprises about 4,000 species distributed in more than 500 genera and 38 families (Javidkar et al. 2015, 2017; Sfenthourakis and Taiti 2015; Dimitriou et al. 2019; Campos-Filho and Taiti 2021; WoRMS 2021).

Terrestrial isopods are one of the most representative taxa in the Brazilian subterranean environments, probably due to the special environmental conditions, such as high humidity, substrate heterogeneity, and absence of specialized predators (Fernandes et al. 2016, 2019). To date, about 210 species are known from Brazil, 70 of which, belonging to the families Styloniscidae, Philosciidae, Scleropactidae, Dubioniscidae, Platyarthridae, Porcellionidae, Armadillidae, Pudeoniscidae, and Armadillidiidae, are recorded from Brazilian caves. Thirty-two taxa are considered to be troglobites (obligatory and restricted to caves), several are troglophiles (facultative cave species), and some species are possibly trogloxenes (Souza et al. 2011; Campos-Filho et al. 2014, 2015a, 2015b, 2016, 2017a, 2017b, 2018, 2019, 2020; Bastos-Pereira et al. 2017; Fernandes et al. 2018, 2019; Cardoso et al. 2020a, 2020b, 2021). The species placed into the last two categories are difficult to assign into a correct subterranean category due to the scarcity of distribution data outside caves.

Among the troglobitic representatives of the Brazilian Oniscidea, 12 species have an amphibious way of life, all in the Styloniscidae: *Chaimowiczia uai* Cardoso, Bastos-Pereira, Souza & Ferreira, 2021, *C. tatus* Cardoso, Bastos-Pereira, Souza & Ferreira, 2021, *Iuiuniscus iuiuensis* Souza, Ferreira & Senna, 2015, *Spelunconiscus castroi* Campos-Filho, Araujo & Taiti, 2014, *Xangoniscus aganju* Campos-Filho, Araujo & Taiti, 2014, *X. ceci* Cardoso, Bastos-Pereira, Souza & Ferreira, 2020, *X. dagua* Cardoso, Bastos-Pereira, Souza & Ferreira, 2020, *X. ibiracatuensis* Cardoso, Bastos-Pereira, Souza & Ferreira, 2020, *X. itacarambiensis* Bastos-Pereira, Souza & Ferreira, 2017, *X. lundi* Cardoso, Bastos-Pereira, Souza & Ferreira, 2020, *X. odara* Campos-Filho, Bichuette & Taiti, 2016, and *X. santinhoi* Cardoso, Bastos-Pereira, Souza & Ferreira, 2020.

The present work describes two new species of *Xangoniscus* from the Serra do Ramalho karst area, state of Bahia, both occurring in special microhabitats, namely the upper aquifers fed by infiltration of water in the limestone rock. Moreover, aspects of their ecology and natural history, as well as conservation remarks about this particular and unique fauna and the region of Serra do Ramalho are given.

Materials and methods

Collections and taxonomy

Specimens of *Xangoniscus* were collected by hand with the aid of tweezers and brushes. Specimens were stored in 70% and 100% ethanol. Descriptions are based on morphological characters with the use of micropreparations in Hoyer's

medium (Anderson 1954). For each new species, the diagnosis, type material, description, etymology, and remarks are given. The *habitus* images were taken with the stereomicroscope model Motic SMZ-168 and the Celestron Microcapture Pro. The photographs were prepared with Adobe Photoshop CC Lite (v. 14.2.1). The appendages were illustrated with the aid of a camera lucida mounted on a CH2 Olympus microscope. The final illustrations were prepared using the software GIMP (v. 2.8) according to the method proposed by Montesanto (2015, 2016).

Maps were produced highlighting the caves where both *Xangoniscus* species occur, as well the hydrological attributes and pressures of economic activities in the region, representing impacts to the cave fauna. The distribution map was constructed with QGIS software (v. 3.18.1) and was edited in PowerPoint Microsoft 365 (v. 2108).

The material is deposited in the scientific collection of cave fauna of the Laboratório de Estudos Subterrâneos (LES), Universidade Federal de São Carlos, São Carlos, Brazil (Curator: Maria E. Bichuette).

Ecological and behavioral study

Both populations were studied in one field trip in October 2020, during the dry-wet season transition. Specimens were sampled *ad libitum* (sensu Altmann 1974; Martin and Bateson 2007). In addition, direct observations of other ecological and behavior data were recorded including reproductive aspects, movement, and microhabitat preferences as well as course estimations of abundance and population density. Microhabitat data included cave zone, type of substrate, depth, water current, and physicochemical variables – pH and temperature.

To assess population parameters (density and size of the population), the individuals were counted along microhabitat inside caves, and measurements (such as length, width, and total area) of the occurrence sites were taken. The population parameters were calculated by dividing the number of individuals observed by area of the occurrence site. Photographs and short videos were taken to document the behavior of both species and the influence of the microhabitat on their distribution in the caves (see Supplementary Online Material).

Study area

Serra do Ramalho karst area, State of Bahia, northeastern Brazil

The Serra do Ramalho karst area, southwestern State of Bahia, is located within the Bambuí geomorphological group, which hosts a high number of caves, mostly without legal protection. Some caves within this karst region extend for more than 15 km, developing huge cave systems (Rubbioli et al. 2019). The Serra do Ramalho karst area comprises the municipalities of Coribe, Feira da Mata, Carinhanha, and Serra do Ramalho, and plateaus are made of limestone belonging to the Bambuí geomorphologic group. Moreover, the limestone in the region forms outcrops that extend parallel to the São Francisco River (Auler et al. 2001;

Rubbioli et al. 2019). According to Köppen criteria, the region has a tropical dry climate (=semiarid) ("Aw"), with annual precipitation of about 640 mm (Beck et al. 2018, 2020). Subterranean waters are mainly exploited for human consumption (Belda et al. 2014; Rubbioli et al. 2019). Surrounding vegetation consists of "Caatinga" composed of mesophytic and xeromorphic forests interspersed with "Cerrado" (savannah-like vegetation). The two new species described herein occur in two caves of the Serra do Ramalho karst area within the São Francisco River basin (Figs 1–3). Figures 2 and 3 show details of the habitat of the new *Xangoniscus* species. Human pressures related to common economic activities in the region (mining and land use), and typical vegetation are shown in Fig. 1. Currently, mining and land use pressures represent a great threat to habitat of cave species in the region.

Results

Family Styloniscidae Vandel, 1952

Genus *Xangoniscus* Campos-Filho, Araujo & Taiti, 2014

Type species. *Xangoniscus aganju* Campos-Filho, Araujo & Taiti, 2014, by original designation.

Xangoniscus lapaensis Campos-Filho, Gallo & Bichuette, sp. nov.

http://zoobank.org/855DC7C6-D196-4B19-8340-18DB358A7277
Figures 1, 2, 4–7

Type locality. Bahia, Feira da Mata, Gruna Boca da Lapa cave (13°56'46.4"S, 44°11'12.0"W).

Type material. ***Holotype*** male (LES 27734), Bahia, Feira da Mata, Gruna Boca da Lapa cave, 15 October 2020, leg. ME Bichuette, DF Torres, JS Gallo, L Senna-Horta & JE Gallão. ***Paratypes*** 1 male (parts in slides), 2 females (LES 27735); 1 male, 3 females (one female with parts in slides) (LES 27736), same data as holotype.

Description. Body length: ♂ 8.0 mm, ♀ 9.3 mm. Body unpigmented, slightly robust, lateral sides slightly convex (Figs 2C, D, 4A–C). Dorsal surface smooth with scattered fringed scale setae (Fig. 5A). Pereonite 1 epimera with distal corners slightly developed frontwards, not surpassing cephalon, posterior corners right-angled; pereonite 2–4 epimera gradually directed backwards, posterior corners progressively more acute; pleon slightly narrower than pereon, pleonites 3–5 epimera well-developed and falciform (Figs 4B, 5C). Cephalon (Figs 4C, 5B) with large quadrangular antennary lobes; vertex with lateral depression to fit antennae when extended backwards, profrons with V-shaped suprantennal line; eyes absent. Telson (Fig. 5C) with well pronounced concave sides and broadly rounded apex. Antennula (Fig. 5D) of three articles, medial and distal articles subequal in length, second article bearing one long seta on inner margin, distal article with two long apical aesthetascs. Antenna (Fig. 5E) with fifth article of peduncle longer than flagellum; flagellum of three articles, first article longest, distal

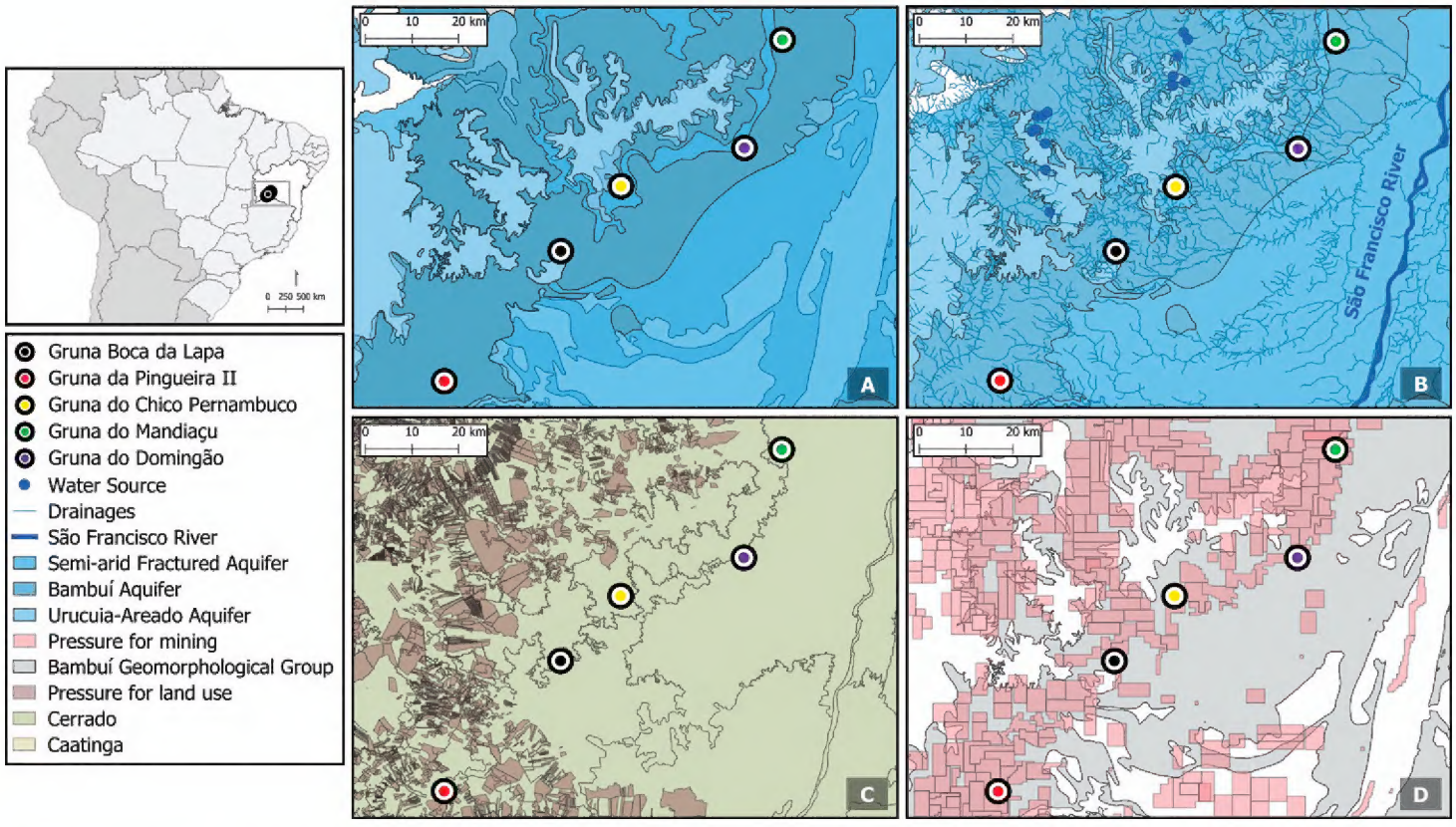

Figure 1. Maps of Serra do Ramalho karst area, State of Bahia, northeastern Brazil **A** limestone outcrops of the Bambuí geomorphological group, Fraturado and Urucuia-Areado aquifer systems showing the type-localities (caves) of *X. lapaensis* sp. nov. and *X. loboi* sp. nov. **B** headwaters and São Francisco river basin **C** pressure for land use showing human-caused alterations **D** pressure for mining activities.

article bearing tuft of free sensilla. Left mandible (Fig. 5F) with two penicils, right mandible (Fig. 5G) with one penicil and lacinia mobilis leaf-shaped with rounded apex. Maxillula (Fig. 5H) outer branch with 5 + 5 teeth, apically entire, and two thick plumose stalks; inner branch with three penicils, proximal one longer than two apical. Maxilla (Fig. 5I) with setose and bilobate apex, outer lobe smaller. Maxilliped (Fig. 5J) basis enlarged on distal portion bearing fringe of fine setae; first article of palp with two setae, distal articles fused bearing distal tufts of several thin setae; endite rectangular, outer and medial margins setose, apically with two triangular teeth and large rounded penicil. Uropod (Fig. 6A) branches inserted at same level, endopod slightly longer than exopod. Pereopods 1–7 gradually elongated, pereopods 1–7 merus, carpus, and propodus bearing sparse setae and fringed scales (discernible over 200× magnification) on sternal margin, pereopod 7 basis with scales of water conduction system; dactylus of one claw bearing many setae on inner and outer margins.

Male. Pereopods 1–6 (Fig. 6B–G) merus with proximal portion bearing fringed scales and thin setae on sternal margin. Pereopod 1 (Fig. 6B) carpus with large antennal grooming brush. Pereopod 5 (Fig. 6F) slightly depressed on distal sternal margin. Pereopod 6 (Fig. 6G) ischium flattened on sternal portion bearing brush of fine setae. Pereopod 7 (Fig. 6H) ischium with straight sternal margin, merus and carpus with no distinct modifications. Genital papilla (Fig. 7A) lanceolate, enlarged on median portion, apical part narrow. Pleopod 1 (Fig. 7B) exopod subtriangular bearing two short setae; endopod longer than exopod, narrow basal article and flagelliform distal article; basipod with triangular medial part fringed with fine and long setae on outer and

Figure 2. Natural habitat and habitus of *Xangoniscus lapaensis* sp. nov. **A** entrance of Gruna Boca da Lapa cave **B** travertine pools where *Xangoniscus lapaensis* sp. nov. occurs **C** specimen of *Xangoniscus lapaensis* sp. nov. submerged on rocky substrate at the travertine pools **D** specimens of *Xangoniscus lapaensis* sp. nov. submerged on silt and rocky substrate.

distal margins, distal margin strongly sinuous. Pleopod 2 (Fig. 7C) exopod triangular, median portion narrower, distal margin almost straight bearing three setae; endopod of two thickset articles, second article about three times as long as first, with complex apical part: distal margin subquadrangular with narrow transverse process, subapical triangular lobe strongly folded. Pleopod 3 exopod (Fig. 7D) subtriangular, longer than wide, covering pleopods 1 and 2, bearing many short setae, distal external margin straight. Pleopod 4 and 5 exopods (Fig. 7E, F) subrectangular, wider than long, with distal portion bearing many short setae.

Etymology. The new species is named after the locality where the specimens were collected, Gruna Boca da Lapa, an important cave discovered and mapped by Grupo

Bambuí de Pesquisas Espeleológicas (GBPE) in 1991 and only now is it studied in relation to its cave fauna.

Remarks. *Xangoniscus lapaensis* sp. nov. resembles *X. aganju* in the shape of the male pleopod 2 endopod; however, it differs in the vertex of the cephalon that lacks a frontal groove, and with a more pronounced suprantennal line that surpasses the antennule insertion (not surpassing in *X. aganju*). Also, the male pereopod 6 carpus lacks any modification (strongly modified in *X. aganju*, see Campos-Filho et al. 2014); the male pleopod 1 exopod is not elongated, and the male pleopod 2 exopod is narrower on inner portion. Moreover, *X. lapaensis* sp. nov. differs in the uropod endopod slightly longer than exopod (opposite in *X. aganju*) and the compact triangular shape of the male pleopod 1 exopod (elongated in *X. aganju*).

Ecology and behavior. *Xangoniscus lapaensis* sp. nov. occurs in travertine pools fed by infiltration of water through the limestone rock, dripping from the cave ceiling, and in isolated pools along the cave (Fig. 2B, C), distributed from the twilight until the aphotic zone, something that demonstrates that its distribution is related to pools formed by infiltration water, i.e., its special habitat is available. The microhabitat is composed of rocky substrate, with little silt and sand sediment. The water is clear, with a very slow current, and a depth varying from 0.05 m to 0.15 m (Fig. 2B–D). Physicochemical variables were typical of karst aquifers that have high pH values: pH = 8.5–9.0, T = 23.5 °C, very different from the conditions in the base-level river (pH = 7.5, T = 25 °C). A small quantity of organic and particulate matter was observed in the small pools. Population density in the pools was high, ranging 6.5 to 32 individuals.m^{-2} and the abundance was 74 individuals in an area of 2.8 m^2. The locomotor behavior was very particular, exhibiting fast and effective walking movements when submerged. However, when emerging from water, individuals showed a slow walking behavior on terrestrial substrate (see Supplementary Online Material 1). *Xangoniscus lapaensis* sp. nov. did not show photophobic behavior (indifferent to flashlight of about 600 lumens), and also seemed to exhibit intraspecific interactions, with no agonistic behavior. Adults and juveniles were observed in the pools, representing possible recruitment at the end of the dry season. It appears that *X. lapaensis* sp. nov. occurs preferentially in microhabitats fed by infiltration water, since no individual was recorded in the stream of the cave, a base-level river. This is very specific and unique, and probably the main habitat of this species is the spaces in the rocks filled by water.

Xangoniscus loboi **Campos-Filho, Gallão & Bichuette, sp. nov.**
http://zoobank.org/0572EC0F-C9B4-4B05-804C-3606AC59A8AF
Figures 1, 3, 8–11

Type locality. Bahia, Feira da Mata, Gruna da Pingueira II cave (14°12'04.3"S, 44°24'39.1"W).

Type material. ***Holotype*** male (LES 27737), Bahia, Feira da Mata, Gruna da Pingueira II cave, 14 October 2020, leg. ME Bichuette, DF Torres, JS Gallo & JE Gallão. ***Para-***

Figure 3. Natural habitat and habitus of *Xangoniscus loboi* sp. nov. **A** karst area where Gruna da Pingueira II cave is located **B** karst and entrance of Gruna da Pingueira II cave **C** specimen of *Xangoniscus loboi* sp. nov. submerged on the silt substrate **D** damn inside the cave **E** specimens of *Xangoniscus loboi* sp. nov. feeding on seed fruits **F** specimens of *Xangoniscus loboi* sp. nov. submerged on silt and rocky substrate **G** specimens of *Xangoniscus loboi* sp. nov. submerged on the rocky substrate **H** stream inside Gruna da Pingueira II cave where *Xangoniscus loboi* sp. nov. occurs.

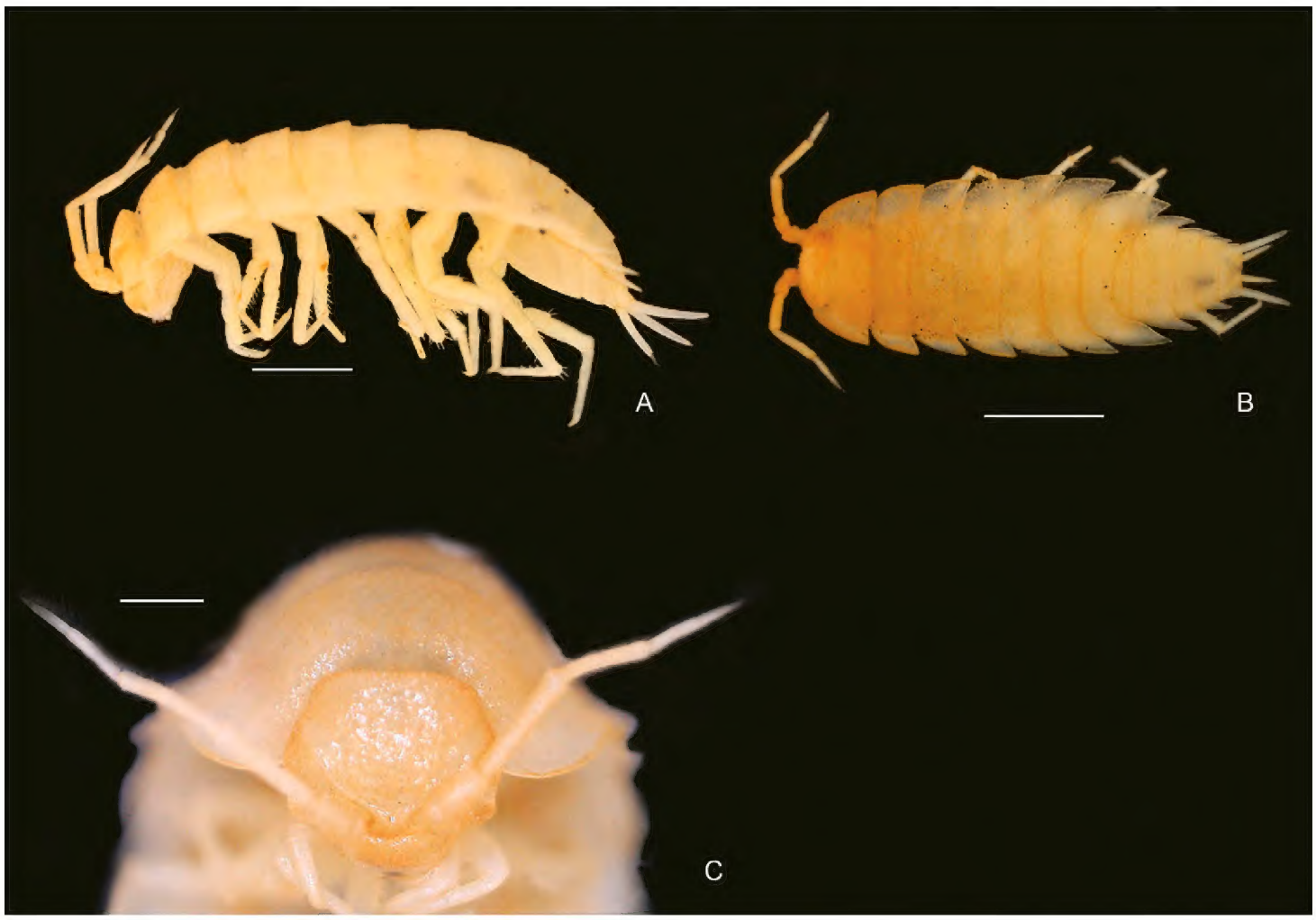

Figure 4. *Xangoniscus lapaensis* sp. nov. **A** habitus, lateral view **B** habitus, dorsal view **C** cephalon and pereonites 1–4, dorsal view. Scale bar: 2 mm (**A, B**); 1 mm (**C**).

types 2 males, 3 females (one female with parts in micropreparations) (LES 27738), 6 males (one male in micropreparations), 4 females (LES 27739), same data as holotype.

Description. Body length: ♂ 12 mm, ♀ 10 mm. Body unpigmented, slender and elongated, lateral sides almost parallel (Figs 3C, E, 8A–C). Dorsal surface smooth with scattered fringed scale setae (Figs 8C, 9A). Pereonite 1 epimera with distal corners not developed frontwards, posterior corners right-angled; pereonites 2–4 epimera gradually directed backwards, posterior corners progressively more acute; pleon slightly narrower than pereon, pleonites 3–5 epimera very short, slightly visible on dorsal view (Figs 8A, 9C). Cephalon (Figs 8D, 9B) with large quadrangular antennary lobes; vertex with slightly lateral depression to fit antennae when extended backwards, profrons with V-shaped suprantennal line, not surpassing antennule insertion; eyes absent. Telson (Fig. 9C) with well pronounced concave sides and broadly rounded apex. Antennula (Fig. 9D) of three articles, proximal and distal articles subequal in length, second article bearing short setae on inner margin, distal article with five aesthetascs. Antenna (Fig. 9E) with fifth article of peduncle longer than flagellum; flagellum of four articles, first article longest, distal article bearing tuft of free sensilla. Left mandible (Fig. 9F) with two penicils, right mandible (Fig. 9G) with one penicil and lacinia mobilis leaf shaped. Maxillula (Fig. 9H) outer branch with 5 + 5 teeth, apically entire, and two thick plumose stalks; inner branch with three penicils, proximal one longer than two apical. Maxilla (Fig. 9I) with setose and bilobate apex, outer lobe smaller bearing one strong

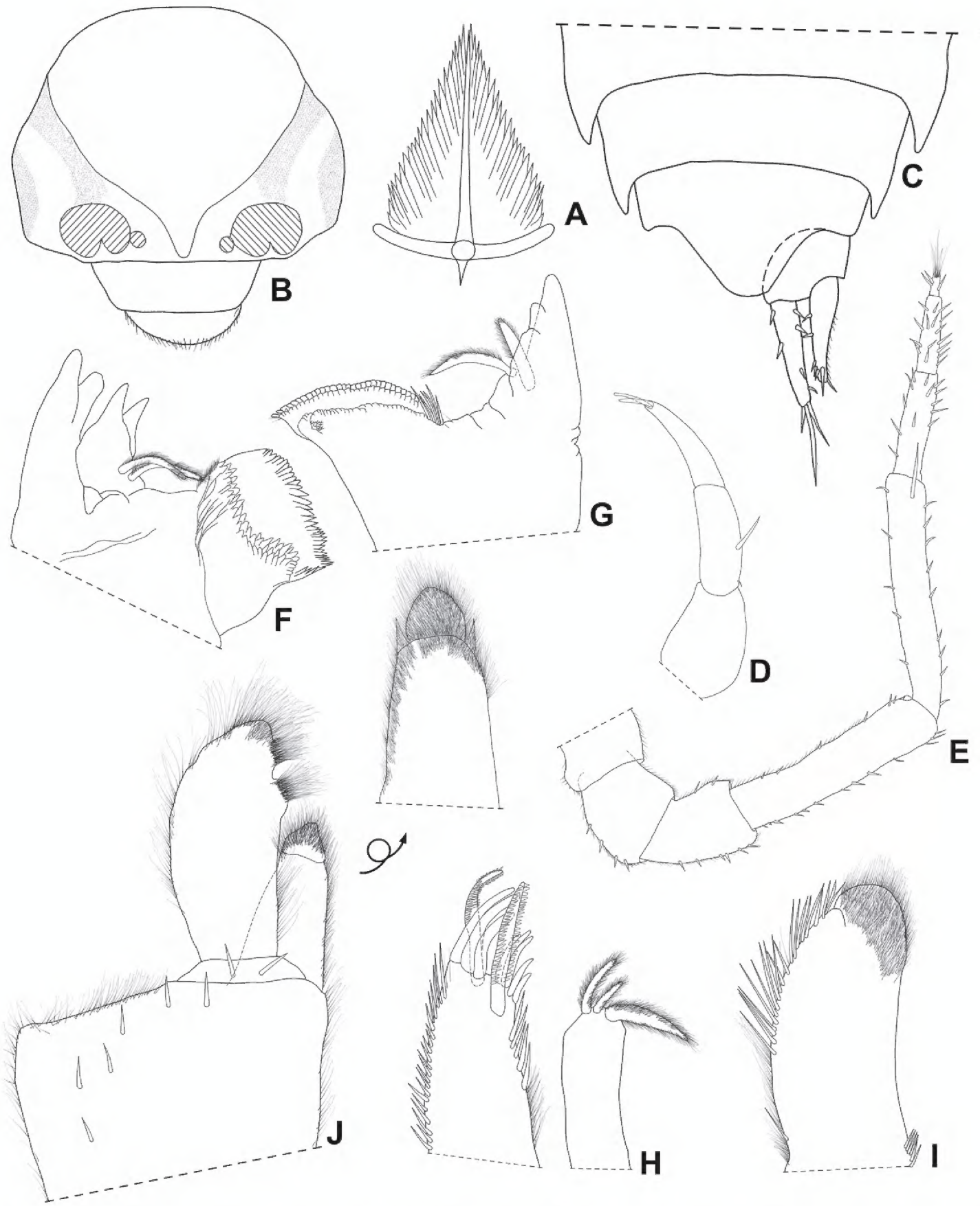

Figure 5. *Xangoniscus lapaensis* sp. nov. Male **A** dorsal scale-setae **C** pleonites 4 and 5, telson and uropod. Female **B** cephalon, frontal view **D** antennula **E** antenna **F** left mandible **G** right mandible **H** maxillula **I** maxilla **J** maxilliped.

seta. Maxilliped (Fig. 9J) basis enlarged on distal portion bearing fringe of fine setae; first article of palp with two setae, distal articles fused bearing distal tufts of fine setae; endite rectangular, outer and medial margins setose, apically with one triangular teeth and large triangular penicil. Uropod (Fig. 10A) branches inserted at same level, endo-

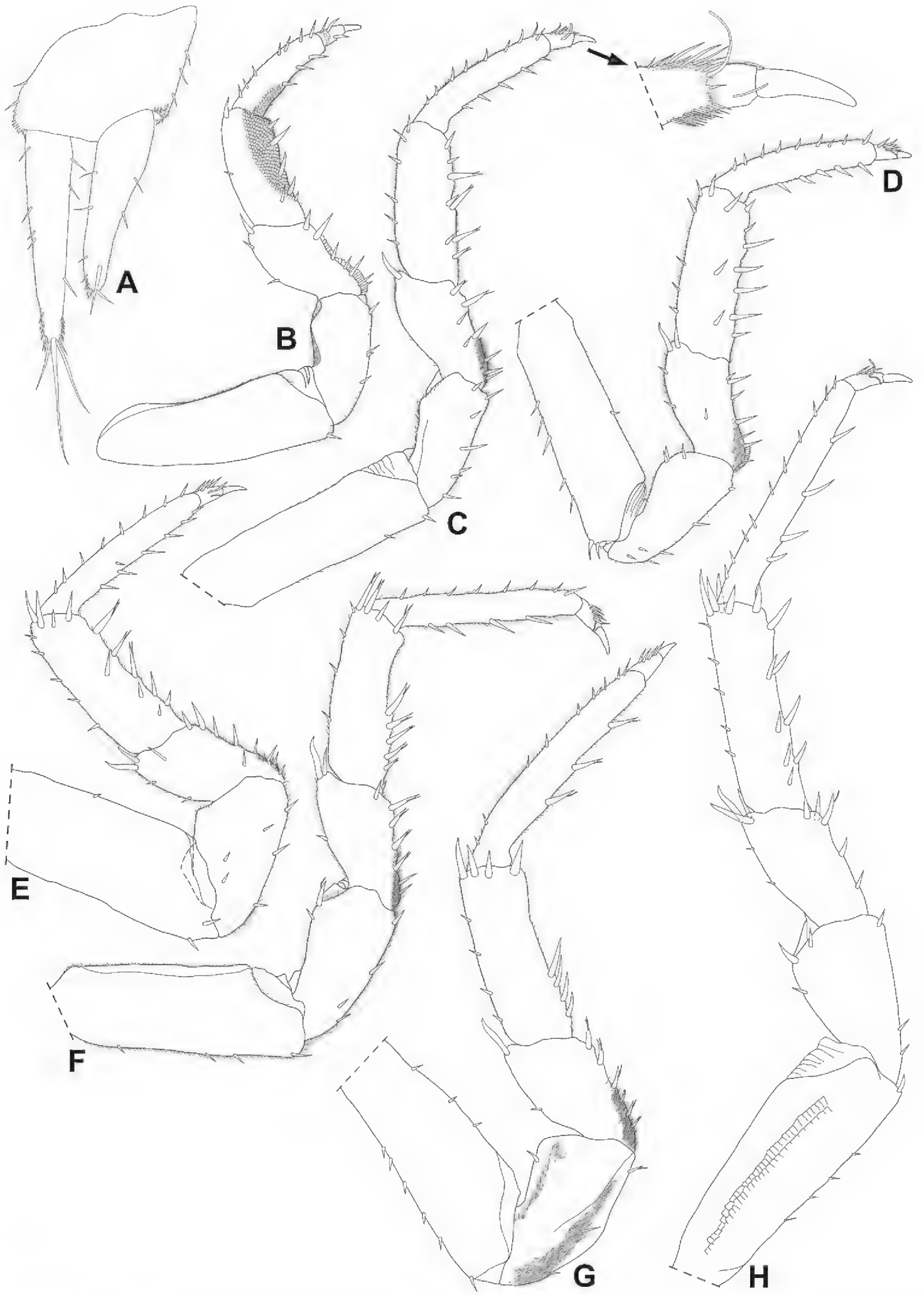

Figure 6. *Xangoniscus lapaensis* sp. nov. Male **A** uropod **B** pereopod 1 **C** pereopod 2 **D** pereopod 3 **E** pereopod 4 **F** pereopod 5 **G** pereopod 6 **H** pereopod 7.

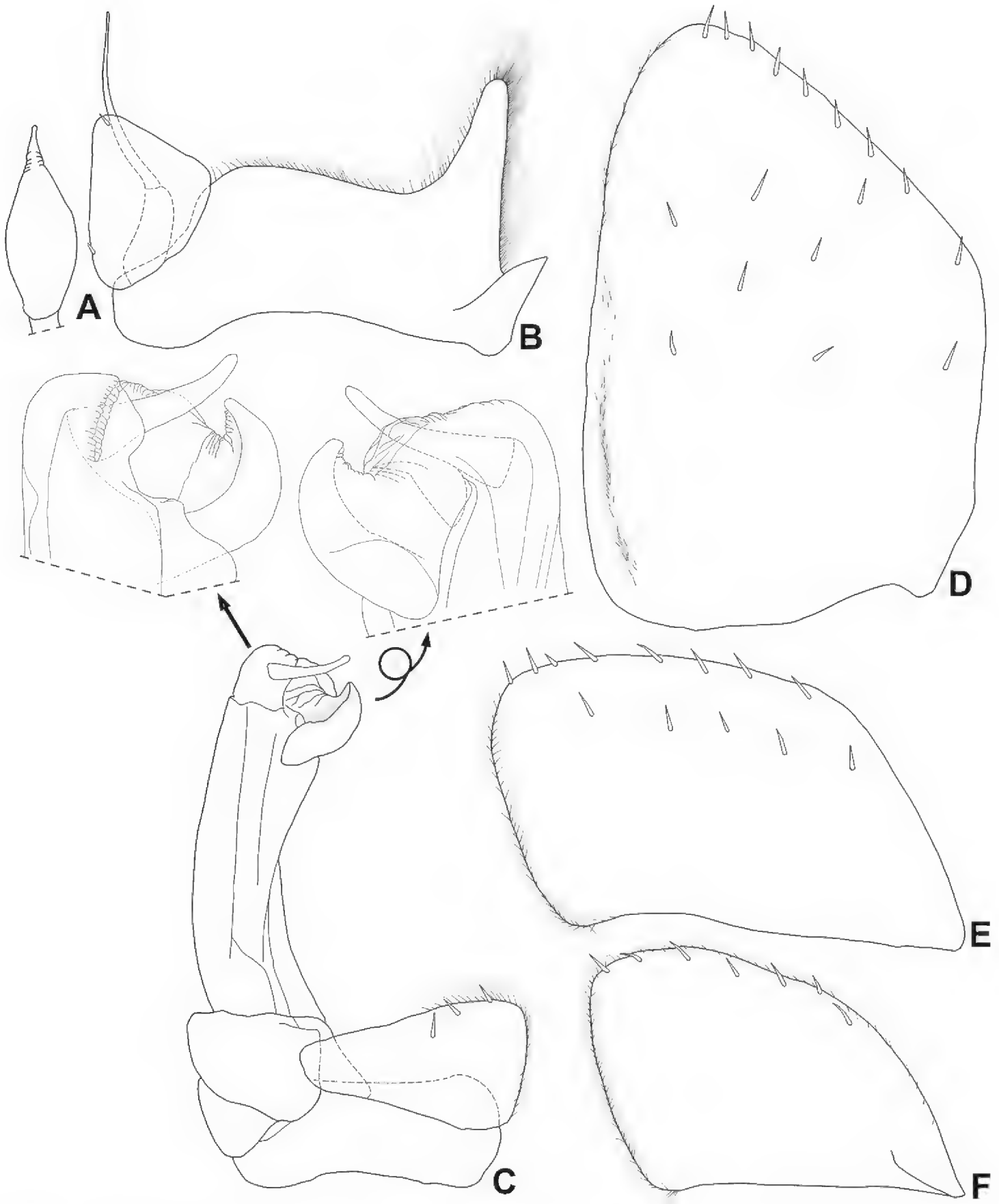

Figure 7. *Xangoniscus lapaensis* sp. nov. Male **A** genital papilla **B** pleopod 1 **C** Pleopod 2 **D** pleopod 3 exopod **E** pleopod 4 exopod **F** pleopod 5 exopod.

pod and exopod subequal in length. Pereopods 1–7 gradually elongated, pereopods 1–7 merus, carpus and propodus bearing sparse setae and fringed scales (discernible over 200× magnification) on sternal margin, pereopod 7 basis with scales of water conduction system; dactylus of one claw bearing many setae on inner and outer margins.

Male. Pereopods 1–3 (Fig. 10B–D) merus with proximal lobe bearing fringed scales and fine setae on sternal margin. Pereopods 4–6 (Fig. 10E-G) merus with dense field of

Figure 8. *Xangoniscus loboi* sp. nov. **A** habitus, lateral view **B** habitus, dorsal view **C** habitus, lateral view **D** cephalon and pereonite 1, frontal view. Scale bar: 2 mm (**A-C**); 1 mm (**D**).

short setae on sternal margin. Pereopod 6 (Fig. 10G) merus bearing brush of setae on sternal margin. Pereopod 7 (Fig. 10H) ischium straight on sternal margin, merus, and carpus with no distinct modifications. Genital papilla (Fig. 11A) lanceolate, enlarged on median portion, apical part narrow and pointed. Pleopod 1 (Fig. 11B) exopod subtriangular, longer than wide, inner and outer margins bearing fringe of fine setae; endopod slightly longer than exopod, narrow basal article and flagelliform distal article; basipod with stout and broadly rounded medial part bearing fringe of fine setae along outer and distal margins. Pleopod 2 (Fig. 11C) exopod triangular, median portion narrower, distal margin almost straight bearing three setae; endopod of two thickset articles, second article about three times as long as first, with complex apical part: distal margin rounded with two narrow transverse processes, subapical circular lobe strongly folded. Pleopod 3 exopod (Fig. 11D) triangular, slightly longer than wide, covering pleopods 1 and 2, bearing many short setae on distal margin. Pleopod 4 exopod (Fig. 11E) subrectangular, wider than long, distal portion bearing many short setae. Pleopod 5 exopod (Fig. 11F) subtrapezoidal, distal margin rounded bearing many short setae.

Etymology. The new species is named after Alexandre Lobo for all his contributions for knowledge and conservation of the Brazilian speleological heritage.

Remarks. *Xangoniscus loboi* sp. nov. differs from all other species of the genus in having the proximal lobe of male pereopods 1–3 merus covered with scales and fine setae, and in the shape of the distal complex apparatus of the male pleopod 2 endopod.

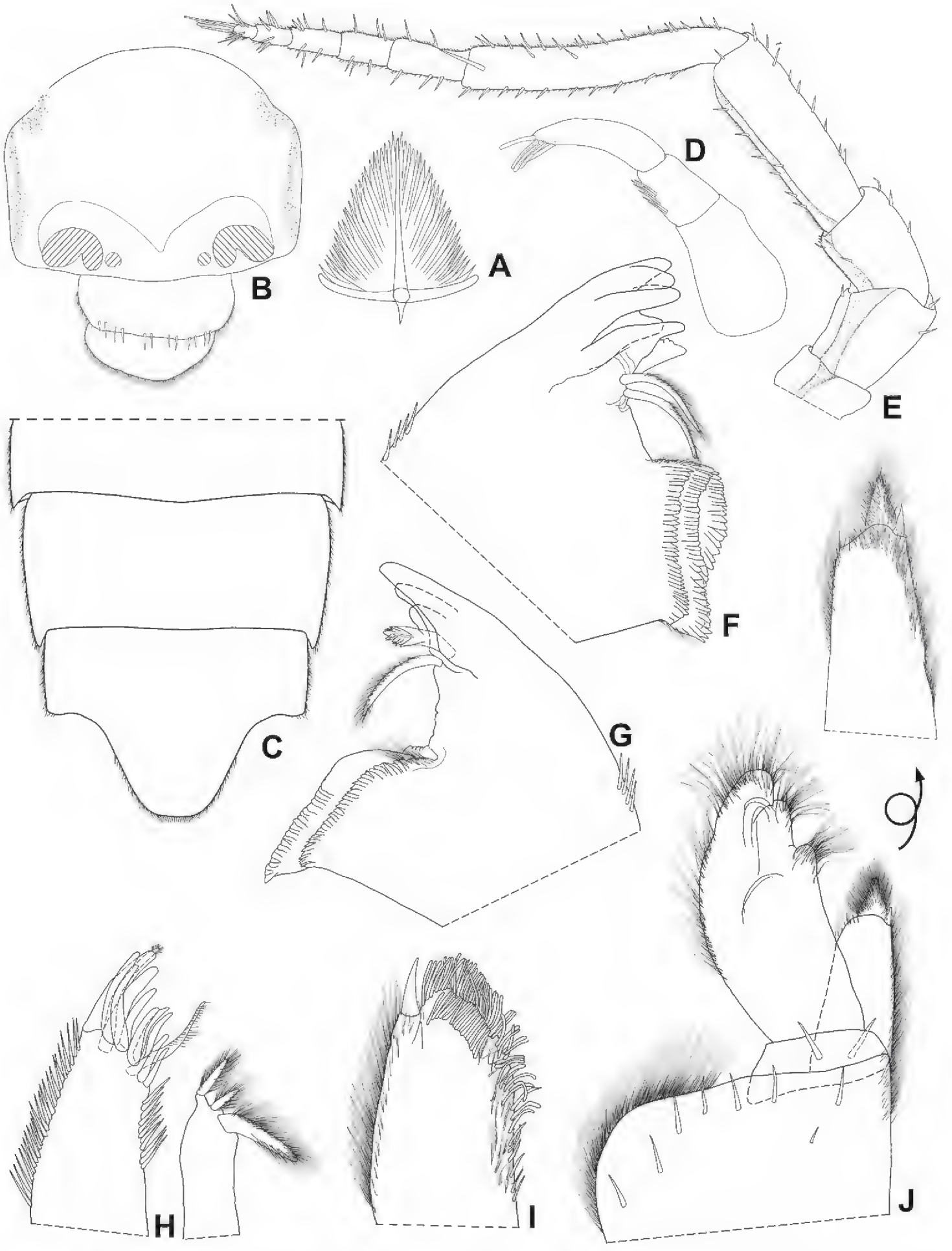

Figure 9. *Xangoniscus loboi* sp. nov. Male **A** dorsal scale-seta **B** cephalon, frontal view **C** pleonites 4, 5 and telson **D** antennula **E** antenna **F** left mandible **G** right mandible **H** maxillula **I** maxilla **J** maxilliped.

Ecology and behavior. *Xangoniscus loboi* sp. nov. occurs in a small stream formed by infiltration water in the limestone rock (Fig. 3). The small stream crosses an extension of about 9 m through the cave entrance and the twilight zones, with

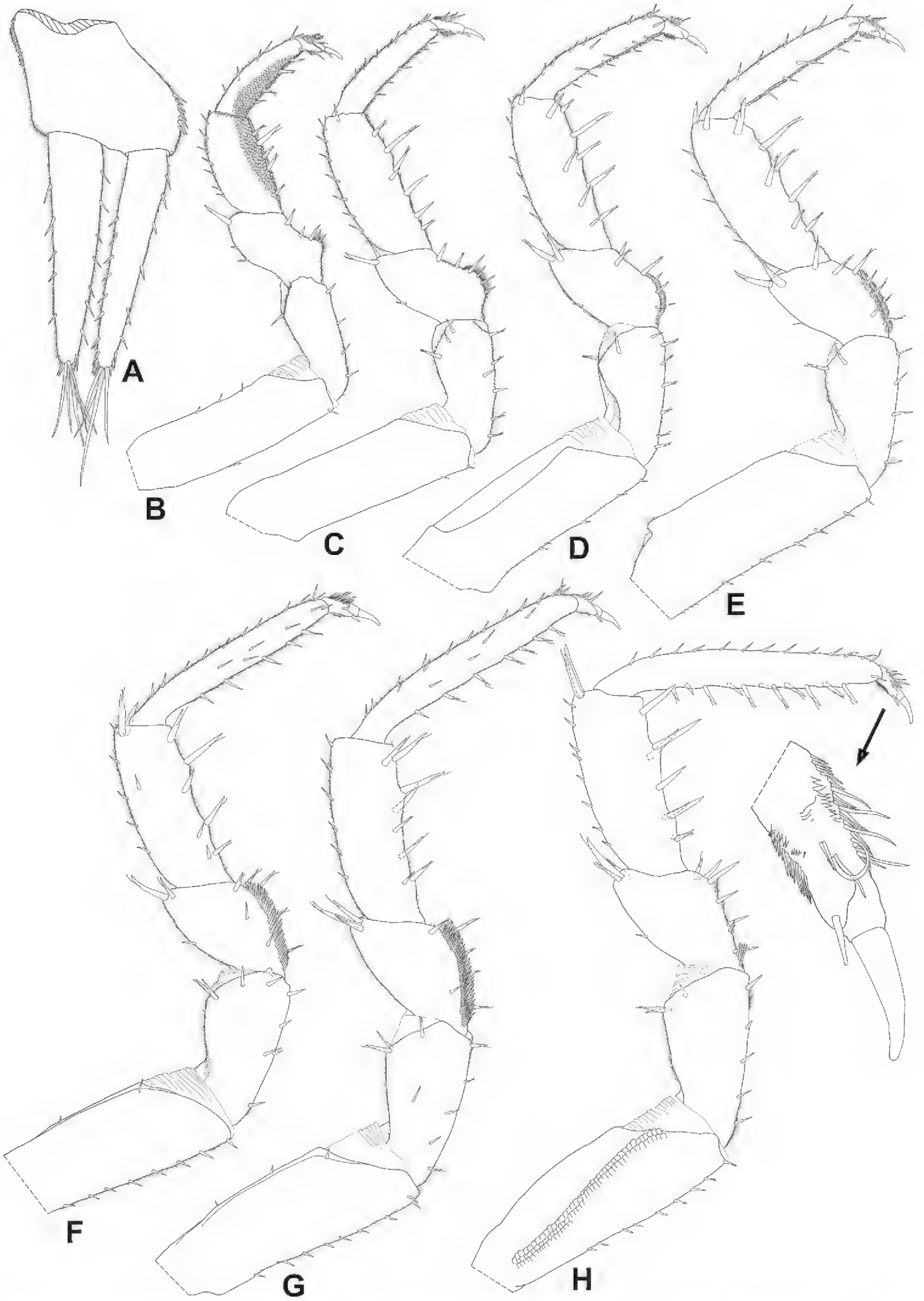

Figure 10. *Xangoniscus loboi* sp. nov. Male **A** uropod **B** pereopod 1 **C** pereopod 2 **D** pereopod 3 **E** pereopod 4 **F** pereopod 5 **G** pereopod 6 **H** pereopod 7.

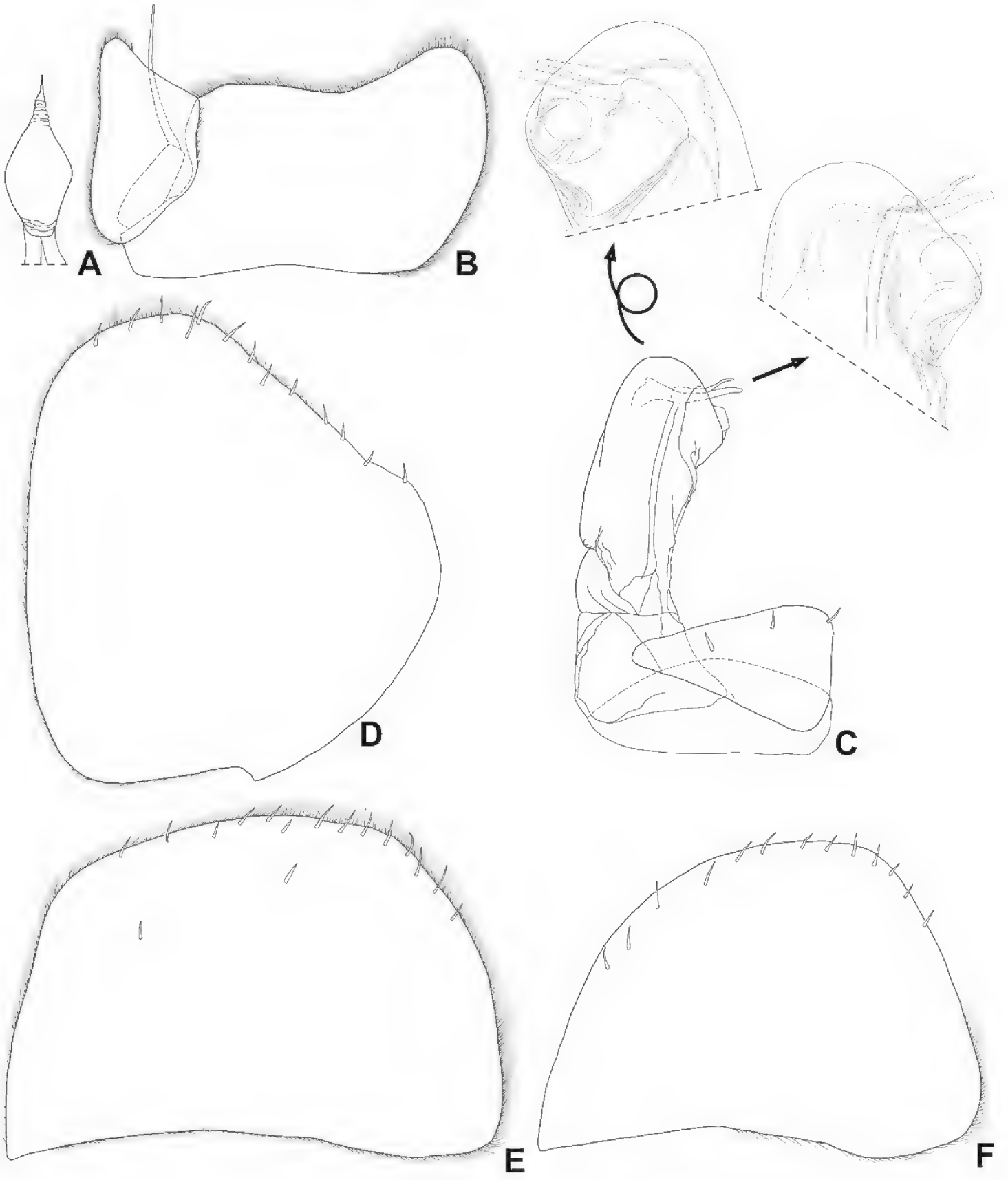

Figure 11. *Xangoniscus loboi* sp. nov. Male **A** genital papilla **B** pleopod 1 **C** Pleopod 2 **D** pleopod 3 exopod **E** pleopod 4 exopod **F** Pleopod 5 exopod.

width varying from 0.2 to 1.2 m and depth from 0.05 to 0.25 m. Along the stream, two small waterfalls (ca. 0.6 m of length) with a rocky substrate were observed. In these stretches, the individuals were seen constantly climbing the substrate (see Supplementary Online Material 2). The bottom of the stream is composed mainly of rocky substrate (concretions and small pebbles), with silt and sand sediment in the bottom, offering rich shelter for the isopods (Fig. 3 C, E–H). The water is clear, with a very slow current. Physicochemical variables are typical of karst aquifers (high

pH values), pH = 8.0–8.5, T = 24.1 °C. A low load of organic and particulate matter was observed in the stream, with some seeds of fruits in the sector close to the cave entrance. The population density in three sectors of the stream were: 54 inds.m^{-2}, 67 inds.m^{-2} and 342 inds.m^{-2}; minimal abundance was 455 individuals in an area of ca. 5.0 m^2. As observed for *X. lapaensis* sp. nov., the locomotor behavior was very particular. *Xangoniscus loboi* sp. nov. shows an efficient walking behavior when submerged (see Supplementary Online Material 3) and climbs vertical stretches (upstream direction) in the stretches with relatively strong water flow. They are clumsy out of water (they do not perform as well as in the submersion behavior), but they adhere very well on dry rock. We also observed copulation: three different couples in reproductive behavior, mated for several minutes (at least 5 min.) (see Supplementary Online Material 4 and 5). Several adults and juvenile individuals were observed along the stream, representing possible recruitment at the end of the dry season. Other interesting behaviors are the indifference to light, even under strong flashlight (ca. 600 lumens), and the interactions among several individuals, touching each other, showing no signs of agonistic behavior. In relation to feeding behavior, some individuals were observed foraging on vegetal debris and seeds (see Supplementary Online Material 6). It appears that *X. loboi* sp. nov. occurs preferentially in microhabitats fed by infiltration water in the limestone. This is very specific and unique, and probably the main habitat of this species is the spaces in rocks filled by water.

Discussion

Both species described herein show the classical signs of troglomorphism (eyes absent and body depigmented), as observed for the other eight valid species of *Xangoniscus*, i.e., *X. aganju*, *X. ceci*, *X. dagua*, *X. ibiracatuensis*, *X. itacarambiensis*, *X. lundi*, *X. odara* and *X. santinhoi*. This could be a pattern for the entire genus, since all species described until now show these same characteristics. Morphological variability in these two characters was not observed for any species of *Xangoniscus*. To date, no *Xangoniscus* species has been recorded outside caves, in the epigean environment, which may denote an ancient isolation of this group.

Morphologically, *X. lapaensis* sp. nov. and *X. aganju* (including specimens from Gruna Chico Pernambuco and Domingão caves, see Campos-Filho et al. 2019) exhibit some level of similarities in the pereopods 5 and 6 of males, e.g., merus and carpus with fringed scales and dense field of thin setae on external margin and carpus distally depressed (see figures of *X. aganju* in Campos-Filho et al. 2014). The complex shape of the distal portion of the male pleopod 2 endopod is quite similar among specimens of *X. aganju* from Gruna do Mandiaçu cave (type locality) and Gruna Chico Pernambuco cave and *X. lapaensis* sp nov. (see figure 13C in Campos-Filho et al. 2014, and figure 7B in Campos-Filho et al. 2019), and specimens of *X. aganju* from Domingão cave and *X. loboi* sp. nov. shares the presence of two transverse processes (see figure 7C in Campos-Filho et al. 2019). Taking into account the geographic distribution of these

species (see Fig. 1) and these morphological characteristics, it is likely that these species represent a species complex. Future phylogenetic analyses potentially will elucidate the hypotheses proposed herein.

The Serra do Ramalho region has been locally exploited historically by agriculture activities and charcoal production on a small scale. At a broader scale, mining projects and large agriculture production along the boundaries of the region pose future threats (Gallão & Bichuette, 2018) (Fig. 1C, D). To date, no effective law guarantees protection of the caves in the region. However, both Gruna Boca da Lapa and Gruna da Pingueira II caves are type-localities of endemic and probably rare troglobites, classifying these caves as maximum relevance *sensu* Brazilian legislation (BRASIL 2008). However, this action likely will not be sufficient, because protection is restricted to a small portion of an extensive massif. The creation of a conservation unit for the entire karst area would probably be the most effective action to ensure the preservation of the biological and speleological heritage of the Serra do Ramalho karst region.

A preliminary evaluation of the conservation status of these species was conducted following the IUCN (International Union of Conservation of Nature) classification. Despite their high population density and abundance, *Xangoniscus lapaensis* sp. nov. and *Xangoniscus loboi* sp. nov. were classified as Critically Endangered (CR) according to the criteria B2ab(iii). Both species are endemic, restricted to their type-locality and found in a specific microhabitat (travertine pools and streams formed by infiltration water). The surrounding areas of the cavities are impacted by deforestation for pastures and the native vegetation is present only at their entrances (Fig. 3A, B). Furthermore, the Gruna Boca da Lapa cave is threatened by uncontrolled tourism. These threats, combined with the impacts mentioned above for the Serra do Ramalho region, justify the conservation status proposed in this evaluation.

Acknowledgements

We thank ICMBio/CECAV (Instituto Chico Mendes para Conservação da Biodiversidade/Centro Nacional de Pesquisa e Conservação de Cavernas) for financial support to the project "Estudos para definição de áreas prioritárias para a Conservação de Proteção Espeleológico na Serra do Ramalho – BA", under execution by the GBPE (Grupo Bambuí de Pesquisas Espeleológicas), coordinated by Ezio Rubbioli and integrated by MEB and LH; to the members of GBPE, E Rubbioli, F Chaimowicz, A Lobo and R Brandi for all support in the occasion of the discovery of the new species; a special thanks to GBPE for sharing speleological information of Serra do Ramalho karst area with MEB and for all contribution to the knowledge of Brazilian caves; to SISBIO (Sistema de Autorização Informação em Biodiversidade) for collection permit to MEB (28992); to Vicerrectoría de Investigaciones of the University of Cartagena for the financial support to the Grupo de Investigación en Biología Descriptiva y Aplicada (Project Redes de Investigación Científica, Acta de Compromiso N° 017–2020); to CAPES (Coordenação de Aperfeiçoamento

de Pessoal de Nível Superior) for financial support to ISC-F during his postdoctoral fellowship (CAPES/PNPD/UFCG/CTRN/PPGRN/201713705) (Finance Code 001); to CNPq (Conselho Nacional de Desenvolvimento Científico e Tecnológico) for productivity fellowship to MEB (310378/2017–6). We would like to thank the anonymous reviewers and editor Dr. ML Niemiller for their critiques and suggestions for the work.

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Supplementary online material

1. Video on YouTube. *Xangoniscus lapaensis* sp. nov. locomotor behavior in the Gruna da Lapa. https://www.youtube.com/watch?v=jFZXOb7BAdg
2. Video on YouTube. *Xangoniscus loboi* sp. nov. moving against the water flow in a small waterfall. https://www.youtube.com/watch?v=ZOwsvT_9gjA
3. Video on YouTube. *Xangoniscus loboi* sp. nov. locomotor behavior in a travertine pool on Gruna da Pingueira II. https://www.youtube.com/watch?v=iuGvySw4SV0
4. Video on YouTube. *Xangoniscus loboi* sp. nov., copula. https://www.youtube.com/watch?v=u2_AK_PfMHs
5. Video on YouTube. *Xangoniscus loboi* sp. nov., copula. https://www.youtube.com/watch?v=yL1PVQHz7OY
6. Video on YouTube. *Xangoniscus loboi* sp. nov., foraging and interacting behaviour. A https://www.youtube.com/watch?v=xQxKHrb80kM